

ORIGINAL ARTICLE

BONE MARROW IN PEDIATRIC PATIENTS WITH HODGKIN'S DISEASE

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Background: Hodgkin's disease is a malignant process of lymphoreticular system that constitutes 6% of childhood cancers. Accurate staging of lymphoma is the basis for rational therapeutic planning and assessment of the presence or absence of marrow involvement is a basic part of the staging evaluation. The objective of this study was to determine the incidence of marrow infiltration in paediatric patients with Hodgkin's disease and to ascertain its morphological spectrum in the marrow. **Methods:** The study included 85 paediatric patients with diagnosed Hodgkin's disease seen at The Children's Hospital/Institute of Child Health, Lahore, from January 2010 to December 2011, referred to haematology department for bone marrow biopsies. **Results:** Ages ranged between two years to fourteen years with an average age of seven years, the male female ratio being 13:1. Mixed cellularity was the commonest histological type present in 66 (78%) cases. The presenting feature common in all cases was superficial lymphadenopathy followed by hepatomegaly in 17 (20%) cases and splenomegaly in 16 (19%). All the marrow aspirates were negative for infiltration, Trephine biopsies revealed marrow infiltration in 9 (10.5%). Five (56%) cases had bilateral while 4 (44%) had unilateral involvement. Pattern of infiltration was diffuse in 8 (89%) and focal in one (11%) trephines. Increased marrow fibrosis was present in eight (89%) cases. Diagnostic Reed Sternberg cells were identified in only one case and the mononuclear variants were present in six cases and atypical cells were present in two cases in these immunohistochemistry for CD15 and CD30 was performed which was positive. Granulomas in one and lymphoid aggregates were present in two trephine biopsies otherwise negative for Hodgkin's infiltration. **Conclusion:** Bone marrow infiltration was present in 10.5% cases, immunohistochemistry was used to confirm infiltration in two cases, the pattern of infiltration being diffuse in majority (89%).

Keywords: Hodgkin's Disease, Bone marrow biopsy

INTRODUCTION

Hodgkin's disease is a malignant process of lymphoreticular system that constitutes 6% of childhood cancers.¹ Hodgkin's disease has a worldwide distribution but shows distinctly different age patterns in different countries. The contrast is greatest between developed and developing countries. Childhood Hodgkin's disease appears to be relatively common and young adult disease relatively rare in developing countries while the reverse is observed in developed regions of the western world.² Hodgkin's disease has a wide age incidence from childhood through to old age.³ The shape of age specific incidence curve for Hodgkin's disease is bimodal with peaks in the decade from 20–30 and in old age, rates for males exceed those for females at all ages.⁴

Approximately 70% patients present with superficial lymphadenopathy involving in order of frequency cervical, auxiliary or inguinal regions.⁵ Mediastinal Hodgkin's disease is found in approximately 10% of patients presentation⁶

Intra-abdominal HD is most commonly represented by involvement of spleen and para-aortic lymph nodes.⁶ Once a histological diagnosis of malignant lymphoma has been established, the next question to be resolved is the extent of the disease,

Accurate staging of lymphoma is the basis for rational therapeutic planning and assessment of the presence or absence of marrow involvement is a basic part of the staging evaluation.⁷

The necessity of bone marrow (BM) biopsy for recognition of Hodgkin's disease involvement is apparent, since diagnostic aspirates are a rarity.⁸ Bone marrow aspirates usually show reactive changes and it is uncommon for neo-plastic cells to be present in an aspirate.⁹ The superiority of BM biopsy over BM aspirate in defining marrow involvement has been well established in Hodgkin's disease.^{10,11} The demonstration of marrow involvement appears to be related to the amount of tissue obtained at biopsy.¹² In this context it has been shown that that bilateral BM biopsies of adequate size are more efficient in detecting lymphoma than unilateral samples.^{10,11}

Histological pattern of marrow involvement in trephine biopsy sections is diffuse in 70–80% of cases and focal in the remainder.¹³ When the pattern is diffuse, Hodgkin's tissue occupies an entire area between bony trabeculae and usually replaces large contiguous areas of marrow, fibrosis is often prominent.¹⁴ When there are only small foci, the trabecular structure is usually not affected, but large areas of involvement are usually accompanied by osteosclerosis, autolysis or osteoporosis of the cancellous bone in their vicinity.¹⁵

In 1971, the committee on Histo-pathological criteria contributing to staging of HL at the Ann Arbor Symposium established the histological criteria for diagnosing BM involvement in patients with proven HL.¹⁶ Incidence of marrow infiltration on diagnosis is reported with a high range of variability in the literature. Marrow involvement mostly results from widely disseminated disease.

The aims of our study were to determine the incidence of marrow infiltration in newly diagnosed paediatric patients and to ascertain the morphologic spectrum of Hodgkin's disease in the marrow.

MATERIAL AND METHODS

This study was carried out in Children's Hospital and Institute of Child Health, Pathology Department, Lahore and included all newly diagnosed biopsy proven paediatric patients of Hodgkin's disease who were referred to the Department of Haematology for BM examination prior to therapy over a period of two years from January 2010 to December 2011.

Patients already diagnosed and on therapy and those presenting with relapse were not included in the study. Clinical data and haematological parameters were collected, and the BM aspirate and biopsy slides were examined.

RESULTS

Eighty-five newly diagnosed cases of HD in children were included in the study; all cases were referred for BM biopsy prior to therapy. There were 79 (93%) males and 6 (7%) females. Ages ranged from 2–14 years, with average age being seven years. There were 72 (84.7%) cases in first decade and 13 (15.3%) in second decade.

Commonest clinical feature was peripheral lymphadenopathy present in all cases followed by hepatomegaly in 17 (20%) and splenomegaly in 16 cases (19%). Anaemia was present in 89% cases with marrow infiltration, the mean Hb being 7.6 gm/dl and in 75% cases without marrow infiltration and mean Hb of 9.3 gm/dl. Leucopenia was present in 67% and 17% cases with and without marrow infiltration with mean total leucocyte count of $4.2 \times 10^3/\mu\text{L}$ and $8.8 \times 10^3/\mu\text{L}$ respectively. Leucocytosis was not present in any of the cases with marrow infiltration while it was present in 8.5% cases without marrow infiltration. Thrombocytopenia was seen in 67% and 20% cases respectively with and without infiltration and mean platelet count being $125 \times 10^3/\mu\text{L}$ and $335 \times 10^3/\mu\text{L}$. Thrombocytosis was not found in any case with involved marrow while it was present in 15.7% cases without marrow infiltration.

Mixed cellularity was the commonest histological type, 66 cases (78%) followed by nodular sclerosis 7 cases (8%). There were 11 (12.9%) unclassified cases as on their lymph node biopsy no

histological subtype of Hodgkin's disease was specified and there was only one case (1.1%) of lymphocyte depletion and none of lymphocyte predominant Hodgkin's disease. Bone marrow aspirate and trephine biopsies were performed bilaterally in 69 cases and unilaterally in 16 cases.

All marrow aspirates were negative for infiltration but reactive changes were noted in 19 (22.3%) cases. Marrow infiltration was found on trephine in 9 (10.5%) cases, with bilateral infiltration in 5 (56%) cases and unilateral infiltration in 4 (44%) cases. In patients with marrow infiltration 89% were males and 11% were female. Seven out of 9 cases with involved marrow were of mixed cellularity type (Table-1) followed by one case each of nodular sclerosis and unclassified type. Diffuse infiltration was present eight cases (89%) and focal in one case only (11%). Fibrosis was observed in eight cases (89%). Typical R-S cells were found in only one case while mononuclear variants were identified in six cases and atypical histiocytes in two cases.

Three trephine biopsies were negative for infiltration but granulomas were present in one while lymphoid aggregates were observed in two trephines.

Table-1: Bone Marrow Infiltration according to Histological Subtypes in Hodgkin's Disease

Histological type	Number of patients	Number of patients with BM involvement	% of patients with BM involvement
Mixed cellularity	66	7	11%
Nodular sclerosis	7	1	14.2%
Lymphocyte depleted	1	0	0%
Lymphocyte-rich predominant	0	0	0%
Unclassified	11	1	9%
TOTAL	85	9	10.5%

DISCUSSION

The diagnosis of Hodgkin's lymphoma is made histologically, usually from an affected lymph node. Once the diagnosis has been confirmed, full staging is carried out by computed tomography (CT) from neck to pelvis, and BM biopsy where indicated. In otherwise localised disease, the pickup rate for upstaging by BM involvement is very low.¹⁷ Bone marrow involvement is detected in less than 2% of children with stage 1A and 11A disease, the reported incidence of BM involvement by Hodgkin's disease in paediatric population at diagnosis ranged from 1.8–6.5%.^{18–20} In our study, the incidence was 10.5%, which is higher than the previous studies but closer to the 9% incidence reported in another study.²¹ In paediatric patients, Bone marrow involvement is associated with higher disease stage.^{18,22,23}

The general criteria for bone marrow

involvement¹⁶ have been previously described and were used as the basis for diagnosis of marrow involvement in this study. Bone marrow aspirates usually show reactive changes. It is uncommon for neo-plastic cells to be present in an aspirate,⁹ smears are invariably non diagnostic for Hodgkin's Disease^{8,24}. In this study also none of the aspirates were positive for Hodgkin's disease, but reactive changes were seen in a few cases.

The highest incidence of BM involvement is found with lymphocyte depletion and mixed cellularity types less than 10% of patients with nodular sclerosis HD have marrow disease at diagnosis and only rare cases of marrow involvement with lymphocyte predominant HD have been reported.^{7,13} Similar findings were noted in this study with highest incidence (78%) of marrow involvement in mixed cellularity type and 11% in nodular sclerosis type, there was only one case of lymphocyte depletion type in this study and marrow involvement was not present in that case. Brunning and Mckenna²⁵ have reported the frequency of BM involvement to be rare in lymphocyte predominance, 5–10% in nodular sclerosis, 20–25% in mixed cellularity and 50–75% in lymphocyte depletion.

Unilateral involvement was present in 4 trephine biopsies (44%) while five (56%) had bilateral involvement. Brunning *et al*¹⁰ reported unilateral involvement with Hodgkin's disease in 43% and bilateral involvement in 57% cases in his study while O'Carroll *et al*¹³ reported unilateral involvement in 2 of 7 positive cases. The size of the biopsy specimen is an important factor in identifying marrow involvement because of the frequent focal nature of HD in the marrow; therefore it would seem reasonable that the yield of positive biopsies, within the limits of stage IV disease, would increase with large specimens.¹³

Marrow involvement occurred in diffuse pattern in the majority, eight cases (89%) and in focal pattern in only one case (11%), similar findings have been reported by O Carroll¹³ they identified focal infiltration in three and diffuse in 12. A lesion was considered diffuse when Hodgkin's tissue occupied the entire area between bony trabeculae and involved large contiguous areas of BM, focal pattern was characterised by small isolated lesions.¹³ Focal infiltration is found in a minority of patients and can show small patchy or nodular lesions surrounded by normal marrow, less frequently a paratrabacular infiltration can be observed, diagnosis of focal infiltration can be missed if the bone marrow biopsy does not have adequate length.²⁶ Only one case (11%) in this study with nodular sclerosis type had focal involvement of the marrow.

Mononuclear variants of RS cells were found in majority of positive trephines while typical RS cells were found in only one case even after prolonged search, Lee *et al*²⁷ also identified RS cells in one of 19 marrow biopsies considered positive for HD, this is in

contrast to the findings of Ocarroll¹³ who found them in majority of cases.

Increased amount of fibrosis to the extent of altering the marrow architecture was present in eight (89%) cases in this study, Myers *et al*²⁸ have reported invariable presence of reticulin or collagen fibrosis associated with involvement of the BM.

Several authors have reported granulomatous lesions in tissues from patients with Hodgkin's disease.^{16,29,30} O Carroll *et al*¹³ identified granulomas in the marrows of 6 patients out of 107 total cases, none of whom had evidence of marrow Hodgkins disease. In the present study granulomas were identified in marrow of one patient who otherwise had no evidence of marrow involvement with Hodgkin's disease.

The presence of atypical histiocytes without features of R-S cells in the cellular background is strongly suggestive of HL, Fibrosis or necrosis alone should be considered suspicious of HL according to the Committee on Histopathological Criteria Contributing to staging of HL at the Ann Arbor Symposium.¹⁶ The diagnostic category of suggestive BM involvement is no longer advisable, as most cases could be interpreted as certain infiltration with the help of immuno-histochemistry.³¹ The immunophenotypic expression of CD30 and CD15, as well as the negativity of LCA, can be adequately exploited for improvement of diagnostic accuracy in cases of suggestive or suspicious diagnosis. In this study marrow infiltration was present in 10.5% cases and the pattern of infiltration being diffuse in the majority 89%. Immunohistochemistry was helpful in confirming suspicion of infiltration in two cases. When adequate immunohistochemical studies are correctly performed and evaluated, certainty of diagnosis can be obtained in almost all cases.

CONCLUSION

Bone marrow infiltration was present in 10.5% cases of Hodgkin's disease. Immunohistochemistry was used to confirm infiltration in two cases, the pattern of infiltration being diffuse in majority (89%).

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